Significance of Conversation between Mast Cells and Nerves

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Abstract

More and more studies are demonstrating interactions between the nervous system and the immune system. However, the functional relevance of this interaction still remains to be elucidated. Such associations have been found in the intestine between nerves and mast cells as well as between eosinophils and plasma cells. Similar morphologic associations have been demonstrated in the liver, mesentery, urinary bladder, and skin. Unmyelinated axons especially were found to associate with mast cells as well as Langerhans' cells in primate as well as murine skin. Although there are several pathways by which immune cells interact with the nervous system, the focus in this review will be on the interaction between mast cells and nerves.

Functional communication between mast cells and nerves has been shown to occur in a variety of both physiologic and pathologic situations.[1,2] Neuronal mechanisms are involved in mast cell activation, and mast cells act as principle transducers of information between peripheral nerves and local inflammatory events. Neuropeptides, released from autonomic or nonadrenergic noncholinergic nerves, may influence the recruitment, proliferation, and activation of leukocytes. On the other hand, inflammatory cells may modulate the neuronal phenotype and function.

Association of Mast Cells and Nerves

It is well established that there is an anatomic association between mast cells and nerves in most

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tissues.[3–6] In various studies, tissue mast cells invariably showed ultrastructural evidence of activation even in normal healthy conditions, suggesting that these cells are constantly providing information to the nervous system. Mutual associations between nerves and mast cells have been observed in normal conditions and in pathologic ones such as human irritable bowel syndrome, atopic dermatitis, and interstitial cystitis.[7] A morphometric study in both infected and healthy rat intestine showed that mast cells and nerves were closely and invariably approximated in rat intestinal villi.[8] Electron microscopy showed evident membrane-membrane association between mucosal mast cells and nerves with dense core vesicles at the points of contact. Other than in the intestine, nerve and mast cell associations are found in rat trachea and peripheral lung tissue,[9] skin,[10] urinary bladder,[11] brain,[12] and several other tissues.[13,14]

Besides an anatomic association, there is a functional bidirectional communication pathway in vivo. For example, psychological stress in rats causes increased chloride ion secretion by the intestinal epithelium, increased colonic mucin

secretion, and increased intestinal permeability, mediated in part by both mast cells and substance P.[15–17] Furthermore, mast cells and substance P–containing nerves are also obligatory components in a hapten-induced model of lung inflammation.[18] Rozniecki and colleagues provided evidence for morphologic, anatomic, and functional interactions of dura mast cells with cholinergic and peptidergic neurons containing substance P and calcitonin gene-related peptide.[19]

Mast Cells

Mast cells are widely distributed throughout the body in both connective tissue and at mucosal surfaces. They form a heterogeneous population of cells with differences in their development, mediator content, and their ability to interact with the local environment.[20] Therefore, it seems likely that mast cells have many diverse functions. They are thought to play a major role in resistance to infection and are extensively involved in inflammation and subsequent tissue repair.[21] Moreover, there is evidence to support the concept that mast cells are functionally important modulators of hair follicle cycling, specifically during anagen development.[22] This invites the exploration of the murine hair cycle as a model for dissecting the physiologic growth modulatory functions of mast cells.[23] Furthermore, mast cells are known to have a significant variety of actions and interactions with other cells and physiologic systems.

Mast cells can be divided into various subpopulations with distinct phenotypes. Mast cell secretory granules contain unique tryptic and chymotryptic serine proteases that differ between species and tissues. The heterogeneity can express itself as differences in histochemical, biochemical, and functional characteristics. The growth factors required for human mast cell differentiation have been shown to be somewhat different than those for such differentiation in rodents.[24] Although tryptase(s) is found in most or every human mast cell, just a single chymase has been defined. Human mast cells are classified by the presence or relative absence of this chymase.[25] In contrast, rodent mast cell subsets store different chymase isoforms. Two main subsets, connective tissue–type mast cells (CTMCs) and mucosal mast cells (MMCs), are recognized as distinct mast cell populations with different phenotypic and functional characteristics.[26,27] Another commonly used classification uses the terms "MCt" and "MCtc"; the MCt phenotype contains tryptase alone whereas the MCtc phenotype contains chymase and tryptase.[28]

In spite of their variation, the different mast-cell subsets are derived from a common precursor in the bone marrow. Mast cell progenitor cells translocate from bone marrow to mucosal and connective tissues to locally undergo differentiation into mature forms. They possess a remarkable degree of plasticity, so that even apparently fully differentiated CTMCs will transform their phenotype to that of MMCs if transplanted into a mucosal environment.[29]

Mast Cell Mediators

Mast cells are capable of the synthesis of a large number of pro- and anti-inflammatory mediators, including cytokines, growth factors and products of arachidonic acid metabolism. Pre-stored mediators, such as histamine, serine proteases, proteoglycans, sulphatases, and tumour necrosis factor (TNF), are released within minutes after degranulation of the cell.[30] After this primary response, a second wave of newly synthesized mediators are released, including prostaglandins and leukotrienes. In the late-phase allergic response, cytokines such as interleukin (IL)-4, IL-5, IL-6, IL-8, IL-13, and TNF are induced and secreted.[30] Expression of this host of cytokines has led to the assumption of a role for mast cells in host defense, for example, in immunoglobulin E (IgE)–dependent immune responses to certain parasites, in natural immunity to bacterial infections, and in inflammatory and allergic diseases.

The communication between mast cells and nerves via cytokines has not received much attention. TNF, which is pre-stored and is released rapidly on degranulation, has an important functional effect. Mast cells also secrete newly

synthesized TNF within 30 minutes following certain stimuli.[31] Furthermore, TNF is able itself to induce mast cell degranulation. TNF is involved in changing neuronal cell function because it can modulate the susceptibility of neurons to electrical stimuli. The sensitizing effect of TNF seems to primarily target C fibres.[32] In vitro incubation of rat sensory nerves with TNF enhanced the response of C fibres to capsaicin.[33] It is known that TNF can activate nerve endings, causing a lowering of the threshold to stimulation. A study by Aranguez and colleagues indicated that mouse astrocytes express TNF receptor 1 (TNFR1).[34] Furthermore, rat microglia transcribe messenger ribonucleic acid (mRNA) for both TNFR1 and TNFR2.[35] These results indicate that neuronal tissue probably expresses both TNF receptors and implies that communication between mast cells and nerves may be mediated, at least in part, by TNF.

Another major mast cell mediator is tryptase, known to be present in all mast cell subtypes. Although proteases (tryptase, chymase) are not classified as cytokines, they have many cytokine-like effects. These cytokine-like activities often activate cells via protease-activated receptors (PARs), cleavage of which results in signal transduction.[36] Proteases regulate neurons and glia in the central nervous system by cleaving PAR. Myenteric neuron protease-activated receptor 2 (PAR2) expression has been detected by reverse transcription polymerase chain reaction. Tryptase has recently been shown to cleave PAR2 on primary spinal afferent neurons, which causes the release of substance P, activation of the neurokinin 1 receptor, and amplification of inflammation and thermal and mechanical hyperalgesia.[37] Corvera and colleagues showed that purified tryptase stimulates calcium mobilization in myenteric neurons.[38] They hypothesized that tryptase excites neurons through PAR2 because activation of PAR2 with trypsin or peptide agonists strongly desensitizes the response to tryptase. In addition, a tryptase inhibitor suppressed calcium mobilization in response to degranulated mast cells. This indicates that tryptase is a major mast cell mediator with the capacity of activating myenteric neurons through PAR2.

Growth Factors

The classic mediators of inflammation are not alone in their ability to influence the interaction between mast cells and nerves. Nerve and mast cell growth factors are thought to play prominent regulatory roles as well. One such factor, nerve growth factor (NGF), acts as a chemoattractant, thereby causing an increase in the number of mast cells as well as their degranulation.[39–41] NGF receptors on mast cells act as autoreceptors, regulating mast cell NGF synthesis and release while at the same time being sensitive to NGF from the environment. Inflammation can lead to an enhanced production and release of NGF. In turn, NGF induces the expression of neuropeptides and lowers the threshold of neurones for firing.[41]

In vivo administration of NGF in neonatal rats caused a great increase in the size and number of mast cells in the peripheral tissues.[42] Furthermore, NGF has been shown to induce degranulation and histamine release from mast cells.[43,44] To complete the circle, mast cells are capable of producing NGF.[45] Therefore, it is not surprising that injection of NGF causes mast cell proliferation, in part by mast cell degranulation.[46]

NGF can have proinflammatory as well as anti-inflammatory effects, depending on the situation and on the concentration of the compound. Braun and colleagues recently showed that nasal treatment of mice with NGF induced airway hyperresponsiveness as measured by electrical field stimulation.[47] Another study by Braun and colleagues showed that nasal treatment of mice with anti-NGF prevented the development of airway hyperresponsiveness.[48] On the other hand, the expression of NGF is increased after brain injury. There is evidence that the increased production of NGF in the central nervous system during brain disease such as multiple sclerosis can suppress inflammation by switching the immune response to an anti-inflammatory suppressive model.[49] In a compelling study, the injection of CD4+ lymphocytes transfected with the *NGF* gene, either before or after the induction of allergic encephalomyelitis, inhibited the onset of demyelination.[50] This powerful inhibition of an autoimmune process showed that local expression

of NGF prevented the migration of inflammatory cells across the epithelium.

Mast Cell Activation by Tachykinins: Expression of the Neurokinin 1 Receptor

In addition to the classic neurotransmitters acetylcholine and noradrenaline, a wide number of peptides with neurotransmitter activity have been identified in the past few decades. Among them, the tachykinins substance P, neurokinin A, and neurokinin B appear to act as mediators of nonadrenergic noncholinergic excitatory neurotransmission.

The tachykinin substance P can activate mast cells via distinct mechanisms. First, substance P can activate mast cells without an intermediary receptor through direct combination with G proteins on the cell surface.[51,52] Second, tachykinins interact with specific membrane proteins belonging to the family of G protein–coupling cell membrane receptors. Three distinct tachykinin receptor subtypes have been identified and are denoted as neurokinin 1 (NK1), neurokinin 2 (NK2), and neurokinin 3 (NK3); these receptors have the highest affinity for substance P, neurokinin A, and neurokinin B, respectively.[53–55] Several investigators have discussed the increased in vivo expression of NK1 receptor in inflamed tissue.[56,57] Therefore, it can be proposed that NK1 receptor expression on immune cells such as mast cells is influenced by environmental inflammatory factors such as cytokines. In previous work, Karimi and colleagues demonstrated the increased sensitivity of bone marrow–derived mast cells (BMMCs) to substance P after a short coculture with the cytokines IL-4 and stem cell factor.[58]

The NK1 receptor appears to be present on the basophil leukemia cell line (RBL).[59] Similar findings were made in rat peritoneal mast cells, which also express NK1 receptors.[60] In an in vitro coculture model, the activation of nerves with scorpion venom elicited the degranulation of RBL cells via substance P.[61] It was shown that this substance-P activation is initiated only at the point of contact between nerve fibres and associated RBL cells through NK1 receptors.[62]

Recently, it has been shown that functional expression of NK1 receptors on BMMCs (which are phenotypically immature mast cells) varies according to culture conditions. The extent of degranulation of BMMCs depends directly on both the concentration of substance P used and the amount of NK1 receptor expression.[63] Similarly, in an in vitro coculture model of BMMCs and neurites, we showed that expression of NK1 by mast cells lowers the threshold of activation induced by nerve stimulation.[64] Furthermore, the response in coculture was inhibited by pretreatment with SR140333, an NK1-specific receptor antagonist strongly pointing to an NK1 receptor–dependent mechanism.

Very recently, Bischoff and colleagues examined the expression of tachykinin receptors on human mast cells and found that human mast cells derived from intestinal mucosa do not constitutively express NK1, NK2, or NK3 receptors.[65] However, when stimulated by IgE receptor crosslinking, these mast cells started to express NK1 receptors but not NK2 or NK3 receptors, again suggesting that specific tissue conditions such as allergic inflammation may lead to mast cell expression of NK1 receptors.

Interaction of Mast Cells and Nerves

Mast cells and nerves are in constant contact with each other in both physiologic and pathologic situations. Many arguments suggest that mast cells and nerves may be seen as a functional unit. They share a number of activating signals, for some of which both cells express receptors (such as vanilloids).[66] Furthermore, both mast cells and nerves respond to stimulation by degranulating preformed mediators, many of which are produced by both cells (NGF, neuropeptides, and endothelin-1). Mast cells can be activated by neuropeptides such as substance P, and many mast cell mediators, including serotonin and tryptase, can cause the release of tachykinins from sensory nerve endings.[3,67–69] Moreover, mast cells and nerves cooperate in a number of pathologic and physiologic processes such as the regulation of hair follicle cycling and development and such as wound

healing.[70,71] Also, stress has been shown to trigger skin mast cell degranulation, an action not only dependent on corticotropin-releasing hormone but apparently also involving substance P.[72] Stimulation of the enteric nervous system by mast cell activation is likely to play an important role in mast cell–mediated host defense in infections, especially infections induced by bacteria.[21,73] Interactions between mast cells and nerves have also been interpreted as important neuronal tissue repair mechanisms following injury.[71,74]

An enhanced interaction between mast cells and nerves can lead to neurogenic inflammation. Inflammatory models have shown a significant increase in the number of mast cells, resulting in the increased release of inflammatory mediators on degranulation. Inflammatory mast cell mediators may modulate sensory nerves through the activation of receptors on nerve terminals (Figure 1). Nonadrenergic non-cholinergic (NANC) nerve endings express receptors for histamine (H1 and H3) and serotonin (5HT2A).[75–77] Under inflammatory-like conditions, primary NANC nerves show an up-regulation of at least histamine H1 receptor expression.[78] A recent report by Shubayev and Myers provides evidence of expression of TNFR1 and TNFR2 in dorsal root ganglia (DRG) neurons in adult rats.[79] Both receptor subtypes were up-regulated in DRG neurons during inflammation. Capsaicin-sensitive nerves can be altered in this way and could result in an increased release of neuropeptides. Allergen/hapten challenge can also lead to production of substance P in a subset of sensory nerve fibres that are typically devoid of neuropeptides. In other words, allergen/hapten challenge leads to a phenotypic switch in the sensory neuropeptide innervation in the airways, probably via mast cell activation, again increasing the interaction between mast cells and substance P–immunoreactive nerves.[80,81] Thus, mast cell activation can result in an increase in the excitability of sensory nerves and the production and secretion of neuropeptides.

Neurogenic Inflammation

Neurogenic inflammation involves a change in function of sensory neurons owing to inflamma-
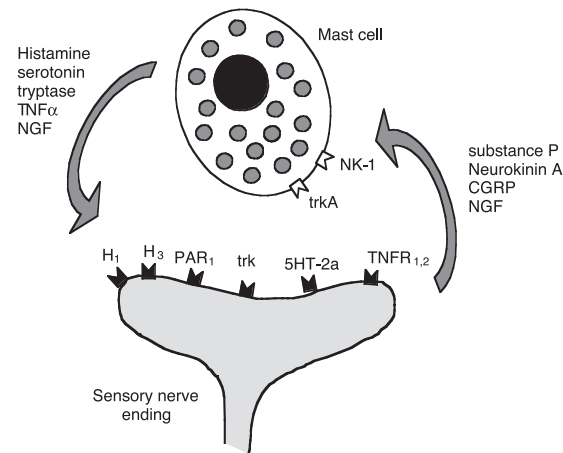

Figure 1 Mast cell–nerve interactions. Inflammatory mediators may modulate sensory nerve endings through the activation of receptors on nerve terminals. Neuropeptides can stimulate mast cells via a receptor-dependent and a receptor-independent mechanism. Under inflammatory-like conditions, receptor expression on nerve endings and mast cells can be up-regulated. CGRP = calcitonin gene–related peptide; H = histamine; 5HT2A = serotonin 2a; NGF = nerve growth factor; NK-1 = neurokinin 1; PAR = protease-activated receptor; TNFR = tumour necrosis factor receptor; trk = neurotrophin tyrosine kinase receptor.

tory mediators, inducing an enhanced release of neuropeptides from the sensory nerve endings.[82] Neurogenic inflammation has been shown to occur in different tissues, including the skin, urinary tract, digestive system, and airways.[83–86] Given the close proximity of mast cells and nerves to blood vessels in most tissues, they may be considered an important functional unit in neurogenic inflammation.[3]

It is becoming apparent that by affecting neuronal functioning, the mast cell and its mediators play an important role in neurogenic inflammation.[3,87] Mast cells pass information on through afferent nerves to local tissues by axon reflexes and to the spinal cord and thence the brain. Stimulation of C fibres by a range of chemical and physical factors results in afferent neuronal conduction that elicits parasympathetic reflexes and antidromic impulses travelling to the peripheral nerve terminal. Axon reflexes account for many of the local physiologic responses to antigen (for instance, in sensitized lung and gut tissues) and have long

been recognized to be involved in local vasodilatation in the skin.[88–91] Antidromic stimulation of guinea pig vagal sensory fibres results in contractions of the isolated airway smooth muscle, mediated by tachykinins.[92] Further studies indicate that neuropeptide release can also be induced via direct depolarization of the nerve terminal.[93]

Priming

It is widely accepted that the effect of substance P as a mast cell secretagogue is found only at high concentrations. However, exposure of mast cells to very small amounts of this neuropeptide may be expected to reduce the threshold of activation of the cells for subsequent challenge with antigen or neuropeptides. Therefore, mast cells can be primed when exposed to physiologically relevant low concentrations of substances, which lowers their thresholds to subsequent activation.

Priming appears to be a broadly based biologic process and has been reported in several cell types. Mast cells have been reported to be primed by different cytokine growth factors for activation by different agonists.[94] Stem cell factor (SCF), for instance, can act as a priming agent in some circumstances.[95] We have shown that SCF and IL-4 prime BMMCs to induce increased responsiveness to substance P.[63] Mast cells can also be primed by substance P itself because repeated doses of very low concentrations (picomolars) of substance P can induce mast cell degranulation and can lower the threshold for degranulation via subsequent crosslinking of IgE receptors by anti-IgE.[96] The concept of priming also applies to neurons. TNF may exert a priming effect (rather than a direct stimulatory effect) on sensory activity.[33,97]

Mast Cell Activation versus Mast Cell Degranulation

Exocytosis is the most obvious event associated with secretion of the mediator molecules contained in granules. It used to be believed that mast cell activation was "all or nothing" and that IgE cross-linking induces the functional consequences of allergic reactions and anaphylaxis. However, the activity of mast cells in health and disease is clearly much more complicated. Secretion can occur without evidence of degranulation, and even molecules stored within the same granules can be released and secreted in a discriminatory pattern.[98]

Mast cells have been increasingly implicated in inflammatory processes in which explosive degranulation is not commonly observed. A study by Ratliff and colleagues ultrastructurally showed mast cells in close proximity to unmyelinated nerve fibres.[99] These mast cells contained granules showing ultrastructural features of activation or piecemeal degranulation, which have been associated with differential secretion. Furthermore, Gottwald and colleagues found increases in the histamine content of intestinal tissues after electrical vagal stimulation without degranulation of mast cells.[100] These data support the potential for intestinal mucosal mast cell regulation by the central nervous system and suggest modulation of mast cells without degranulation. Furthermore, IL-1 stimulates secretion of IL-6 without release of the granule-associated protease tryptase.[101] Selective secretion of IL-6 from mast cells appears to be distinct from degranulation and may contribute to the development of inflammation, in which the importance of IL-6 has been recognized. Serotonin can be released separately from histamine, and differential synthesis and release of arachidonic acid metabolites, prostaglandins, and leukotrienes have been reported.[102,103]

Interaction of Mast Cells and Nerves in Tissues

Brain and Immune System

The brain and the nervous and immune systems are the major adaptive systems of the body.[104] Several pathways have been shown to link the brain and the immune system, such as (1) the autonomic nervous system via direct neural influences and (2) the neuroendocrine humoral outflow via the pituitary. Corticotropin-releasing hormone (CRH), secreted by the pituitary gland, is a major regulator of the hypothalamic-pituitary-adrenal

(HPA) axis and cortisone synthesis and acts as a coordinator of the stress response.[105] CRH is also thought to be involved peripherally in tissue responses to stress in the skin, respiratory tract, and intestine.

Mast cells are resident in the brain of many species.[106] They appear to enter the brain via penetrating blood vessels. Brain mast cells are associated with blood vessels throughout the brain and especially in the meninges.[107] They seem to be involved in behavioural activity, such as the courting behaviour of doves.[108] Large numbers of tryptase-containing mast cells have been described as surrounding the pituitary gland and are thought to act as an immune gate for HPA axis activity.[109] These mast cells can respond to antigens and regulate CRH secretion via histamine effects.[105]

The physiologic significance of mast cells in brain function and/or metabolism is unclear. However, they can modulate neuroendocrine control systems,[2] and they could play a role in the regulation of meningeal blood flow and vessel permeability.[110] Pavlovian conditioning has also been shown to be able to promote mast cell degranulation through as yet unknown mechanisms.[111]

Apart from their being resident cells, mast cells can move through the brain in the absence of inflammation. Mast cells in the central nervous system may participate in the regulation of inflammatory responses through interactions with the HPA axis. Matsumoto and colleagues showed that in the dog, degranulation of mast cells evoked HPA activation in response to histamine release.[109] The physiologic effects of psychological stress are often largely mediated by CRH, released either centrally or peripherally, and mast cell–nerve interactions are important components of this response.[112] In response to psychological stress or certain physical stressors, an inflammatory process may occur through the release of neuropeptides (especially substance P) from sensory nerves and the activation of mast cells or other inflammatory cells. Central neuropeptides initiate a systemic stress response by activation of neuroendocrine pathways (such as the sympathetic nervous system, the hypothalamic-pituitary axis, and the renin-angiotensin system) with the release of stress hormones (ie, catecholamines, corticosteroids, growth hormone, glucagons, and renin).[113] These effects have been found in a variety of stress models, including cold, restraint stress, and water avoidance stress.[15,114,115]

The Skin

The dermis is richly innervated by primary efferent sensory nerves, postganglionic cholinergic parasympathetic nerves, and postganglionic adrenergic and cholinergic sympathetic nerves.[116] Neuropeptides, released by cutaneous nerves, have been shown to activate a number of target cells, including Langerhans' cells, endothelial cells, and mast cells.[117] In the skin, neuropeptides are released in response to nociceptive stimulation by pain and by mechanical and chemical irritants, to mediate skin responses to infection, injury, and wound healing.[118,119] Substance P is one of the main neuropeptides responsible for the skin reaction characterized by erythema, pain, and swelling.[119] In addition, substance P can cause the release of histamine[120] and TNF[121] from skin mast cells, which in turn leads to vasodilation.

Interestingly, capsaicin (which releases neuropeptides from nerves) applied to human skin induces the release of chymase within 6 hours and the induction of E-selectin in adjacent microvascular endothelium, events consistent with release of substance P from axons and subsequent stimulation of cytokine-mediated mast cell interaction with endothelial cells. However, an identical application of capsaicin to human skin grafted onto immunodeficient mice (and thus experimentally lacking in unmyelinated axons) failed to yield similar findings.[5] These results indicate that unmyelinated axons connect Langerhans' cells and dermal mast cells.

Recent studies have suggested that mast cells play a crucial role in the down-regulation of immune responses and the induction of tolerance after exposure of skin to ultraviolet B radiation (UVB). Hart and colleagues reported the involvement of histamine in UVB-induced suppression in mice, and mast cells have been shown to be the source of UVB-induced histamine.[122,123] Furthermore, interactions between mast cells and the nervous system appear to be involved in UVB-mediated immune suppression. TNF, reported to

be derived from mast cells, is a major cytokine implicated in signalling the immunosuppressive effects of UVB.[124] Evidence indicates that mast cells are triggered to release TNF in response to the neuropeptide calcitonin gene–related peptide (CGRP), which is released from UVB-damaged cutaneous nerve endings.[125]

Airways

Efferent and afferent autonomic nerves regulate many aspects of human and animal airway function. In addition to cholinergic and adrenergic innervation, the NANC nervous system is an important third neural network in the lung. Inhibitory NANC nerves contain vasoactive intestinal peptide (VIP) and nitric oxide, which are potent relaxants of the airways and which counteract bronchoconstriction.

Excitatory NANC nerves or so-called sensory nerves are mainly localized in and beneath the airway epithelium. Tachykinins and CGRP are the predominant excitatory NANC neuropeptides in the airways.[126]

Mast cells lining the mucosal layer of the respiratory tract have been found in close proximity to substance P-immunoreactive and CGRP-immunoreactive nerves of rat trachea and peripheral lung tissue.[10] Immunohistochemical studies of neuronal tachykinins in the airways of asthmatic patients have yielded conflicting results. Whereas an increase in both the number and length of tachykinin-immunoreactive nerve fibres in the airways was found in some studies, other studies detected significantly less substance P–like immunoreactivity in lung tissue from asthmatic patients as compared to nonasthmatic patients.[127–130] However, this latter finding may reflect an augmented release of substance P followed by degradation. Studies on autopsy tissue,[130] plasma levels,[131] lung lavage fluid,[128] and sputum[132] suggest that tachykinins are present in increased amounts in asthmatic airways.

Neuropeptides influence the recruitment, proliferation, and activation of inflammatory cells such as mast cells. There is growing evidence that tachykinins and CGRP are involved in neurogenic inflammation of the airways. Structural studies show that mast cells associate with nerves in the lung. Furthermore, Forsythe and colleagues have demonstrated that substance P and neurokinin A induce histamine release from human airway mast cells.[133] Moreover, antigen causes a secretory response in the rat trachea via an interaction dependent on mast cells and nerves.[89]

Gastrointestinal Tract

The gastrointestinal tract is characterized by a unique accumulation of immune and inflammatory cells. The mechanism of interaction between nerve and inflammatory cells in the intestine is, however, very unclear. Intestinal mast cells have been repeatedly reported to communicate with the enteric nervous system. Furthermore, Stead and colleagues, on the basis of electron microscopy studies, reported an anatomic association between mast cells and nerves in the human intestinal mucosa.[134]

Nerve stimulation has been reported to cause mast cell degranulation in the intestine. First, Shanahan and colleagues showed that substance P caused mediator release from intestinal mucosal mast cells.[135] Subsequently, substance P and CGRP fibres have been reported to activate peptidergic mast cells in the intestinal mucosa of healthy and infected rats as well as in patients with inflammatory bowel disease.[1]

Mast cell mediators also appear to have an effect on the nerves in the intestine. Intestinal mast cell infiltration may perturb nerve function, leading to abdominal pain perception in patients with irritable bowel syndrome (IBS).[136] Recent evidence for activated mast cells associated with enteric nerves in IBS strongly implies that mast cells are involved in this symptom complex.[136] A study by Jiang and colleagues using an intestinal model for anaphylaxis showed that serotonin and histamine, released from the mast cells after intestinal anaphylaxis, stimulate mesenteric afferents via 5-HT3 and histamine H1 receptors.[137] Mesenteric afferent-nerve discharge increased approximately 1 minute after luminal antigen challenge and was attenuated by serotonin and histamine receptor antagonists. Mast cell–nerve association appears to function as a homeostatic unit in the regulation of gut physiology and in response to antigens.[138]

Perdue and colleagues determined the existence of an integral nerve-to-mast cell and mast cell-to-nerve connection during intestinal anaphylaxis.[139] A role for the mast cell-to-nerve connection was established by increases in the short-circuit current after antigen challenge. The response to antigenic stimulation was reduced in mast cell–deficient W/Wv mice as compared to their +/+ litter mates and was inhibited by different mast cell antagonists in +/+ mice but not in W/Wv mice, pointing to a mast cell-to-nerve connection. Furthermore, reconstitution of the mast cell deficiency was followed by a restoration of the neural response. In sensitized guinea pig intestine, the short-circuit-current secretory response to antigen occurred simultaneously with acetylcholine release and could be blocked by atropine.[140] This showed conclusively that nerve excitation and the secretion of the main cholinergic neurotransmitter could be induced by antigen via mast cells through an immune-mediated response. The effects of *Clostridium difficile* toxin on intestinal segments has also been shown to be dependent on intact mast cells and substance P–containing nerves.[141,142]

It can be reasonably concluded that nerves and mast cells form a physiologic unit that presumably maintains and regulates homeostasis of the mucosal epithelial secretory response. This unit is involved in health, in response to stress, and also in response to injuries and environmental pathogens.

Therapy

In different tissues and species, there is constant communication between mast cells and the nervous system. This functional communication has been shown to occur in a variety of both physiologic and pathologic situations.[6] The concept of these interactions is very interesting and may bring about new therapeutic and diagnostic approaches.

In humans, an inhaled long-acting β_2 agonist inhibits mast cell mediator release and plasma exudation and may reduce sensory nerve activation. In combination with a corticosteroid, the low systemic effect of these drugs does not result in any significant adverse effects, and there is a strong scientific rationale for long-term asthma therapy.[143] In the skin, cyclosporin A has powerful therapeutic effects on severe therapy-resistant atopic dermatitis.[144] Treating the skin with cyclosporin A increases the stable granule population and results in the disappearance of the close interrelation of mast cells and cutaneous nerves. These findings suggest that cyclosporin A may exert its therapeutic effect by inhibiting mast cell activation and by affecting the interaction between mast cells and nerves.

Exogenous administration of neuropeptides to maintain normal immune defences represents a new field of pharmacotherapeutics against bacterial invasion. But besides this positive health effect of neuropeptides, there is the negative fact that neuropeptides can activate mast cells and result in an enhanced communication between mast cells and nerves, causing an inflammatory response. Mast cell mediators can sensitize sensory neurons, which further activate the mast cells by releasing neurotransmitters or neuropeptides (eg, neurotensin, somatostatin, substance P, and acetylcholine). It has been shown that in the gastrointestinal tract, CGRP, substance P, and VIP-immunoreactive nerve fibres are involved in protection of the tissue.[145,146] In a rat colitis model, an early decrease in these neuropeptides may be an essential condition for the development of colitis. That the intensity and density of substance P and VIP-IR nerve fibres increased after the induction of colitis suggests their possible involvement in tissue repair.[147] Again, on the other hand, these neuropeptides can activate mast cells that play a pivotal role in inflammation. An enhanced interaction between mast cells and nerves can also lead to neurogenic inflammation.

From everything we know so far of the association between mast cells and nerves, it is becoming clearer that the interaction is involved in the regulation of physiologic processes as well as in disease mechanisms. First, therapeutic targets have to be very selective. Because these associations of mast cells and nerves seem to appear throughout the body, it may be very difficult to find a drug that is selectively effective at a particular site in the body. Second, if a selective drug that

provides protection against disease is found, interference in the cross-communication between mast cells and nerves also increases the risk of changing the healthy balance that is essential for maintaining tissue homeostasis.

More physiologic studies are needed for a better understanding of how the activation of mast cells and nerves is modulated, how sensory nerves control mast cell functions, how mast cells use sensory nerves in inducing inflammation, and the role of nerve fibres and their mediators. New findings will continue to increase our understanding of mast cell–nerve associations and their function in health and disease and will be followed by new therapeutic and diagnostic approaches.

Conclusions

Extensive crosstalk exists between nerves and mast cells. Although differences in species have been reported, morphologic as well as functional associations are found in most tissues in humans and in rodents. Many of these associations have been shown to occur between substance P- and CGRP-containing neurons and mast cells of all subtypes.

The role of this bidirectional communication between mast cells and nerves appears to be multifactorial. Mast cells are thought to play a major role in resistance to infection and are extensively involved in inflammation and subsequent tissue repair. The communication with the nervous system allows the peripheral and central nervous systems to be involved in the regulation of defence mechanisms, inflammation, and response to infection. The involvement of mast cell–nerve communication in the response to stress, for instance, points to an extensive communication between the nervous and immune systems.

However, the complexity of the picture has increased further as it has become clear that classic neurotransmitters such as acetylcholine and neuropeptides are produced by nonneuronal cells. Nonneuronal cells of the immune system, such as monocytes, macrophages, T lymphocytes, and eosinophils, have been shown to produce endogenous substance P.[148,149] This alternative source of immune cells could represent an additional source of tachykinins in inflamed tissues, providing a nonneurogenic tachykininergic contribution to the local inflammatory process.[150]

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